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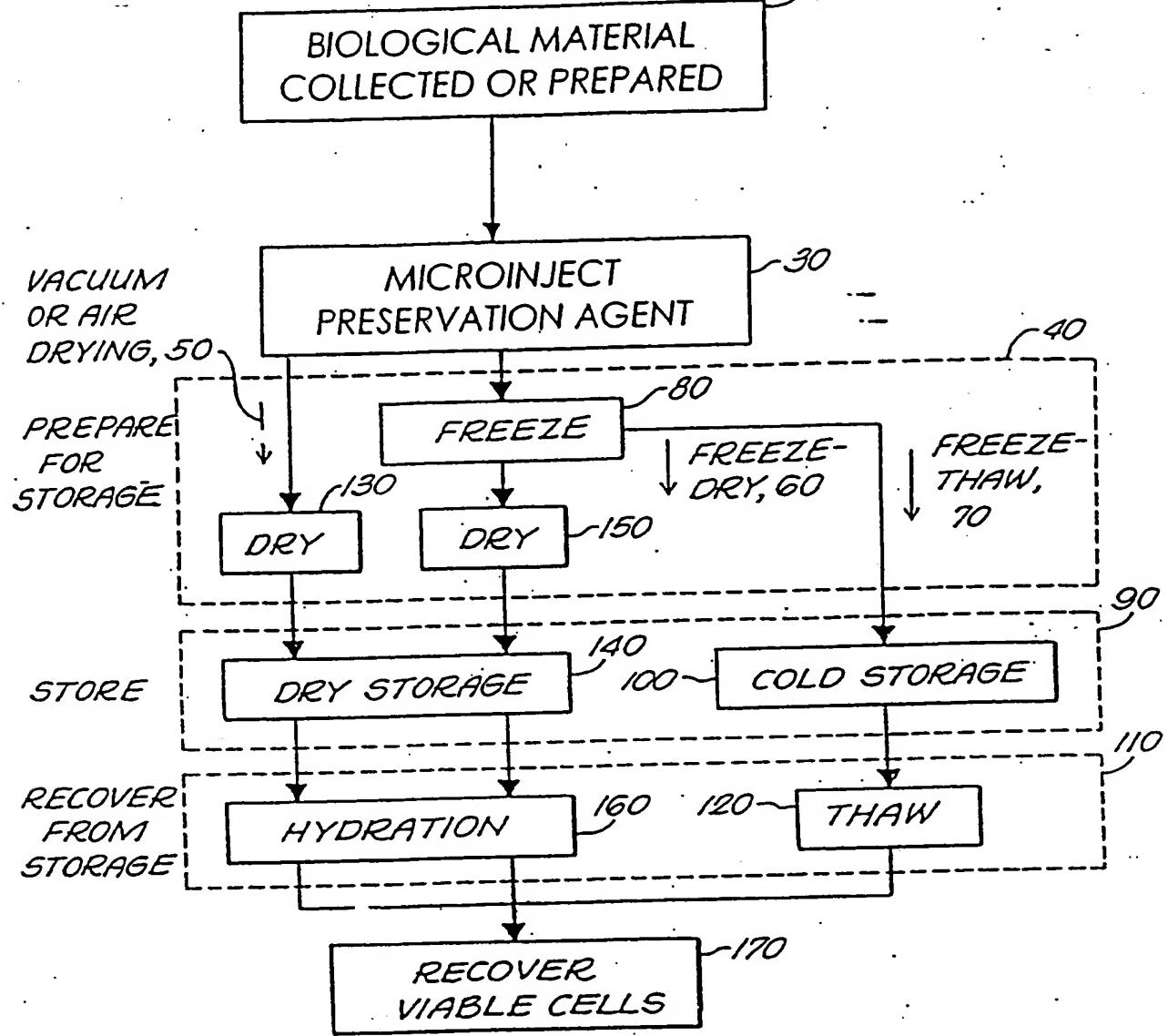


Figure 1

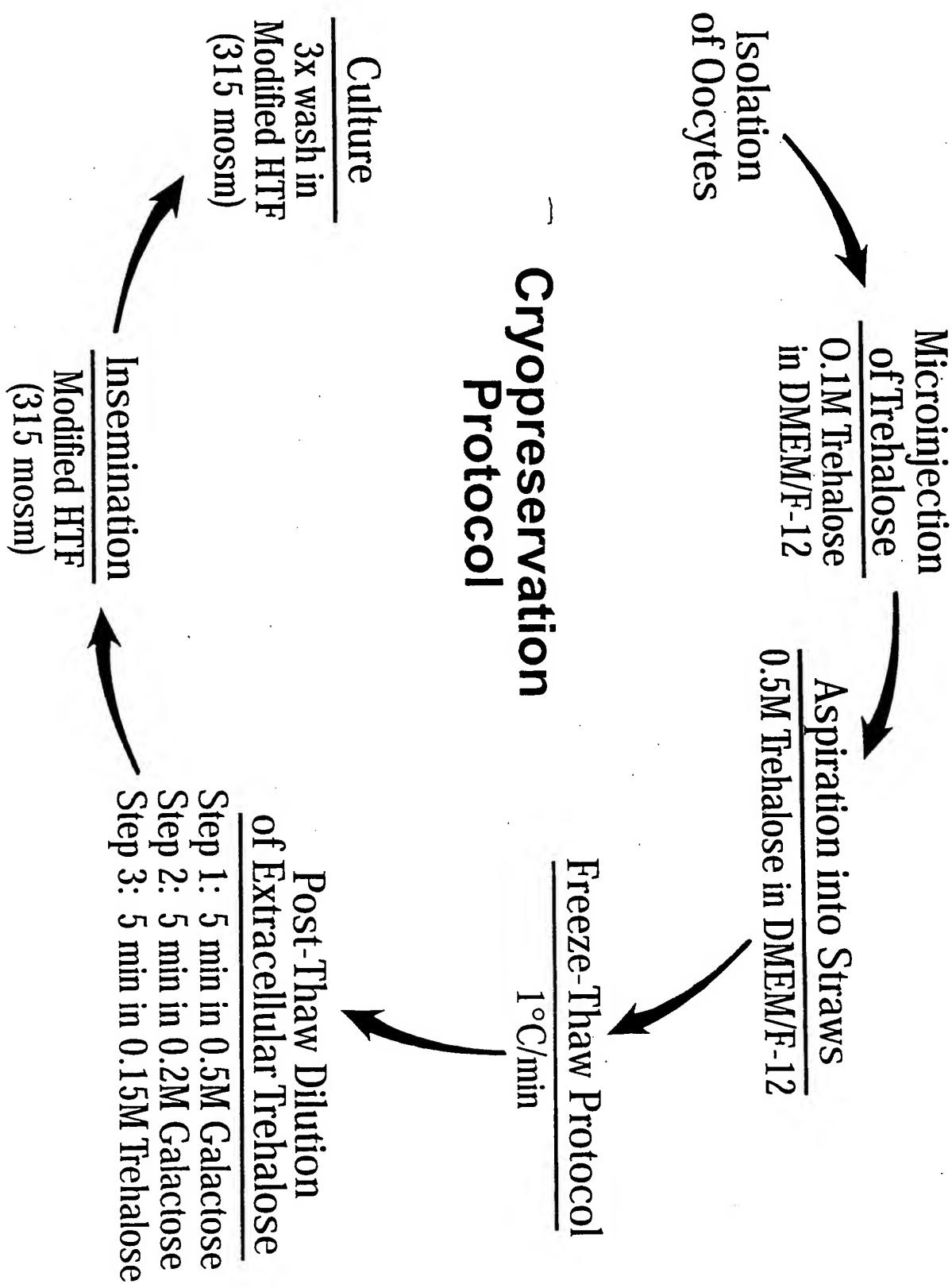
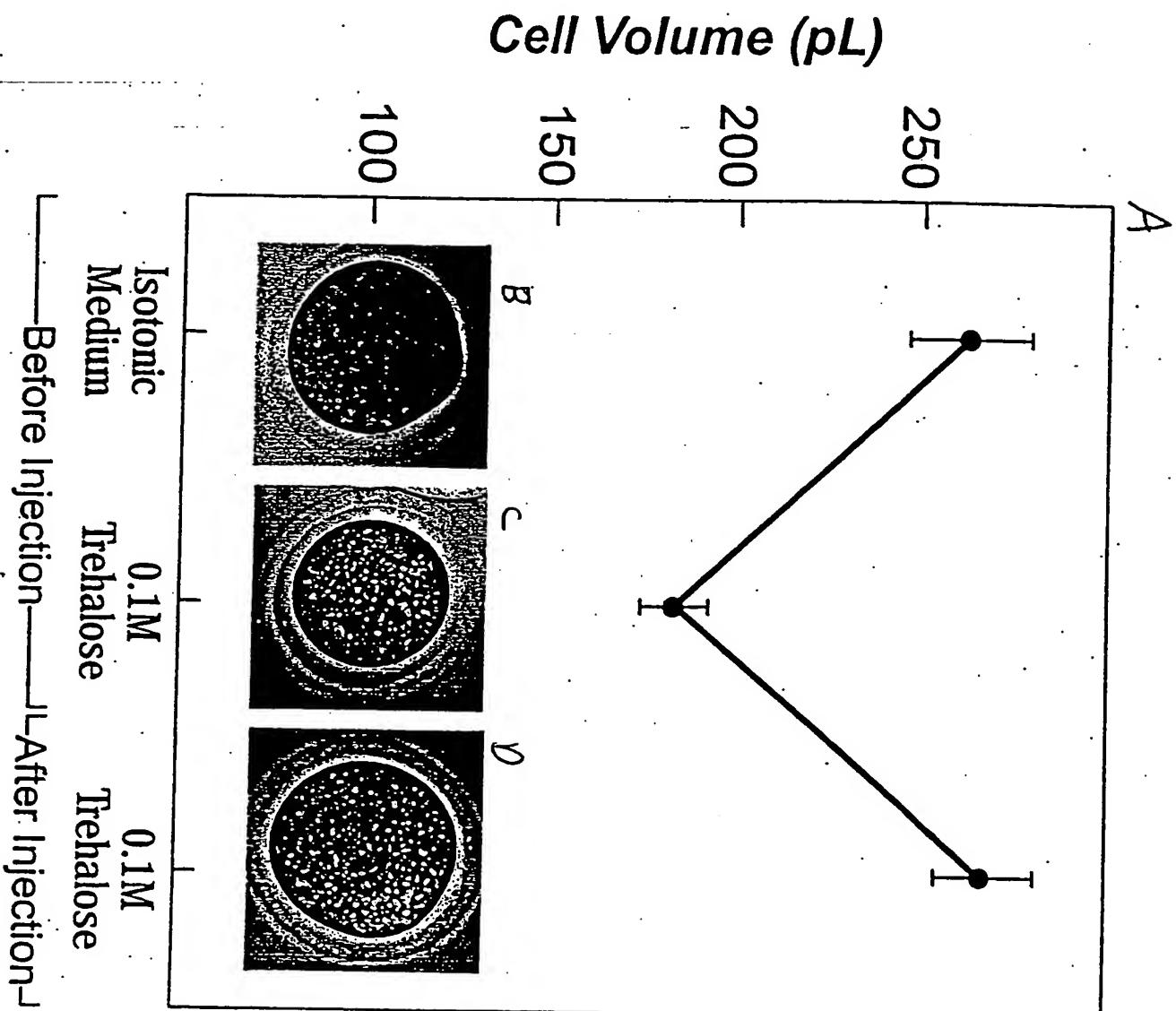
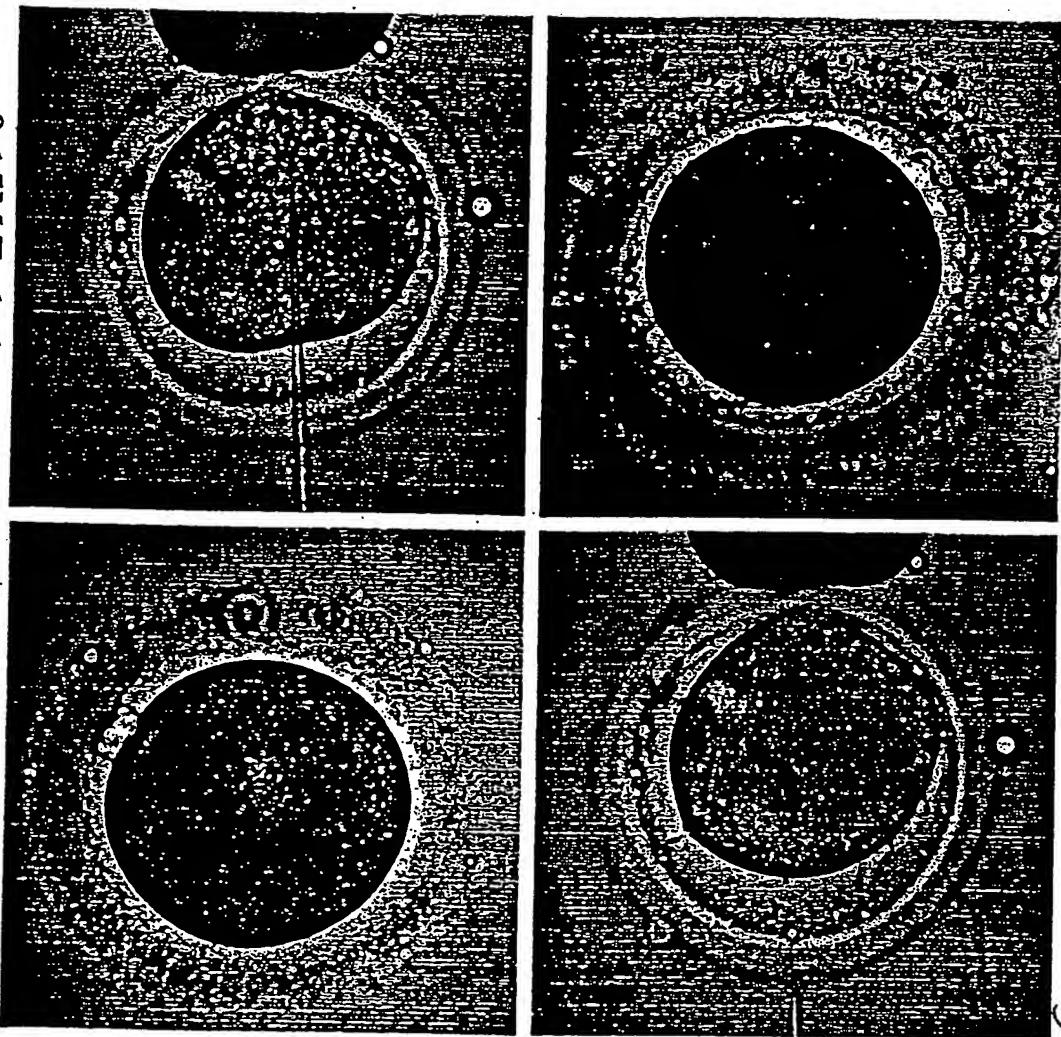


Figure 2



0.15M Trehalose
During Injection 0.15M Trehalose
After Injection



Isotonic Medium
Before Injection

0.15M Trehalose
Before Injection

Figures 4A – 4D

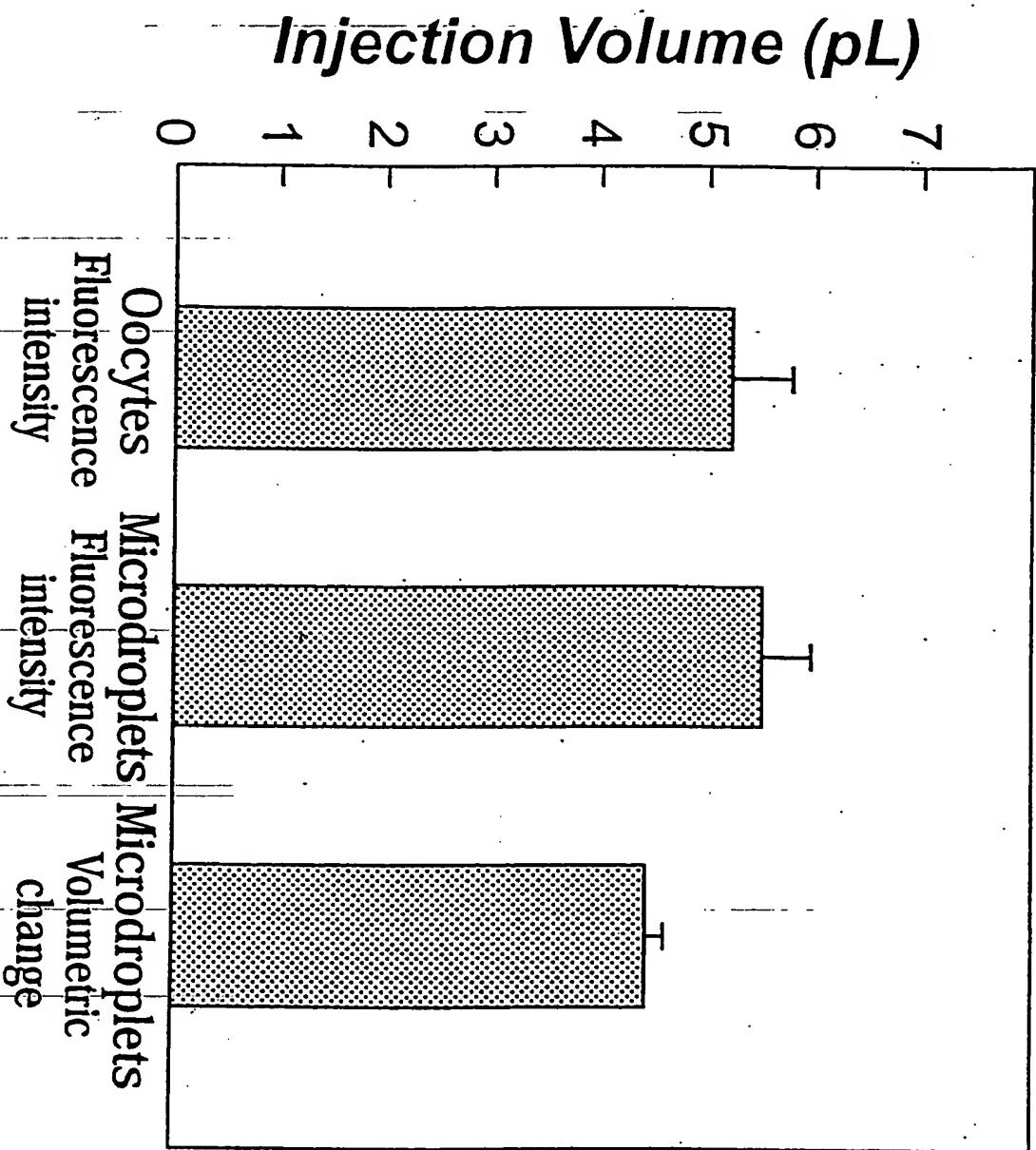
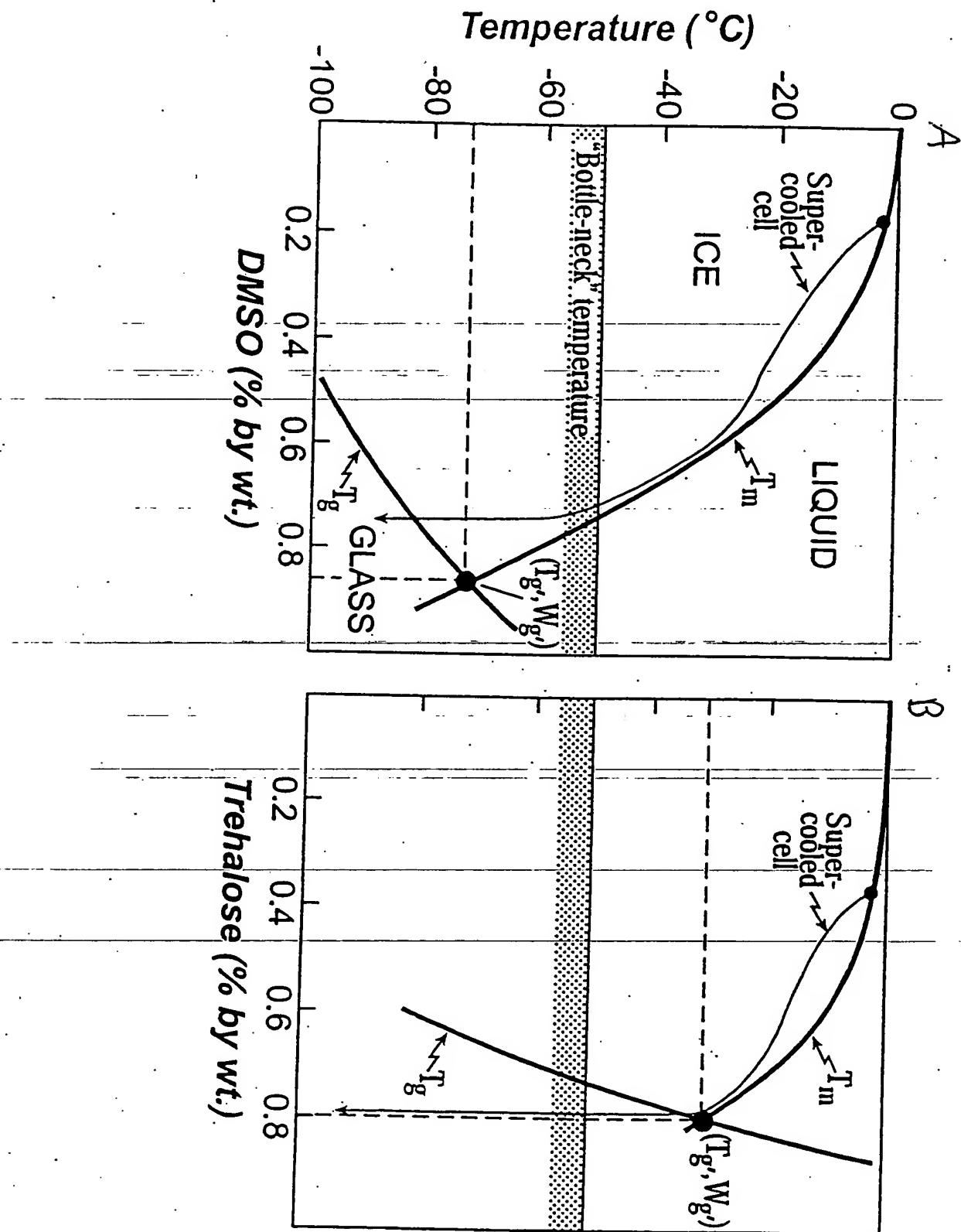


Figure 5



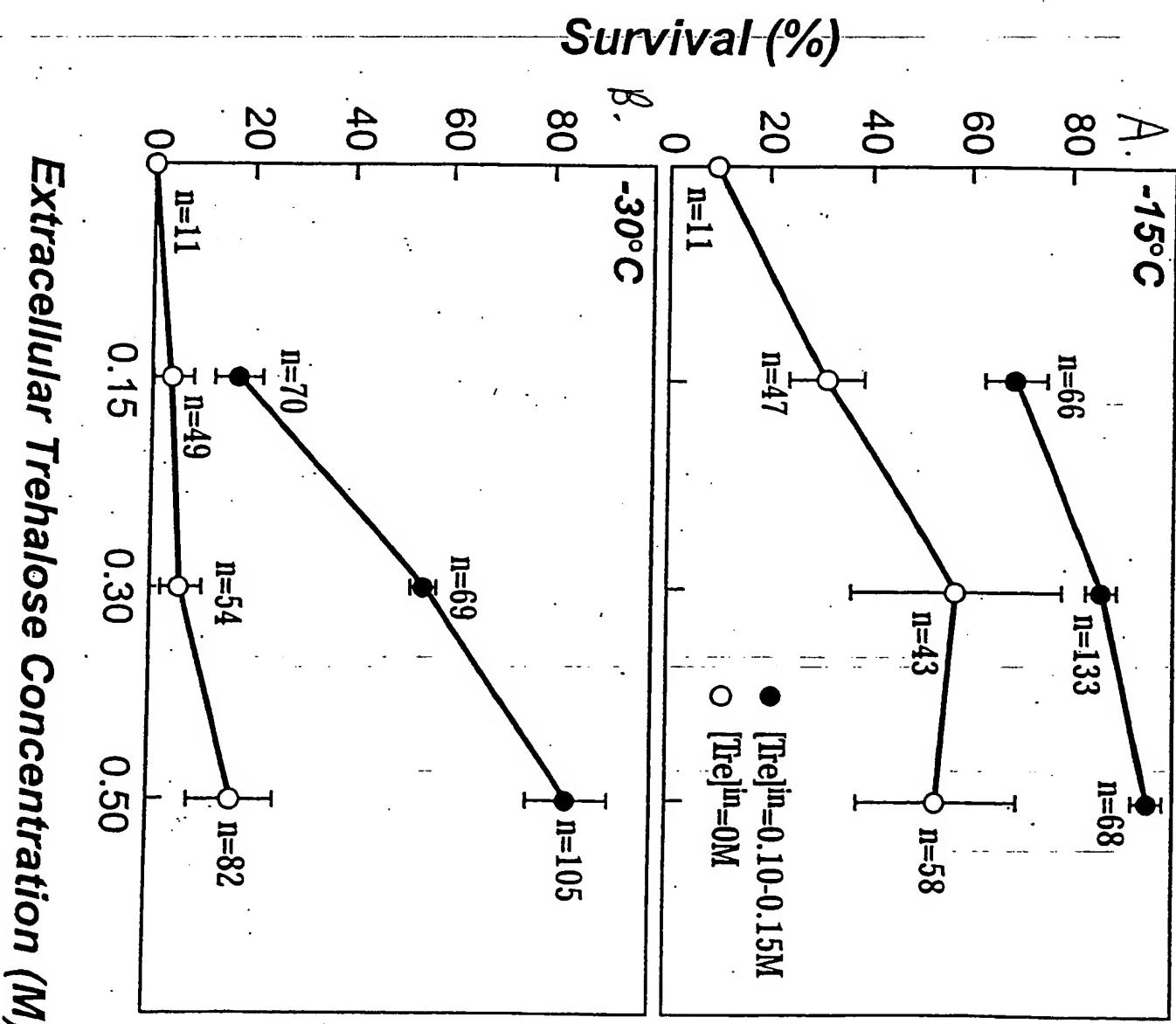
Figures 6A – 6B

Component	HTF	Isotonic	Modified HTF	Hypertonic
NaCl (mM)	101.60	95.00	106.70	
KCl (mM)	4.69	4.78	5.37	
KH ₂ PO ₄ (mM)	0.37	0.38	0.43	
MgSO ₄ • 7H ₂ O (mM)	0.20	0.20	0.22	
NaHCO ₃ (mM)	25.00	25.00	28.07	
CaCl ₂ • 2H ₂ O (mM)	2.04	2.00	2.25	
Lactate (mM)	21.40	20.00	22.46	
Pyruvate (mM)	0.33	0.33	0.37	
D- Glucose (mM)	2.78	2.78	3.12	
L-Glutamine (mM)	—	1.00	1.12	
BSA (mg/ml)	4	4.00	4.00	
EDTA	—	0.01	0.01	
Essential amino acids	—	—	—	
Non-essential amino acids	—	—	—	
Phenol red (% w/v)	0.001	0.001	0.5X	0.5X
Gentamicin (mg/ml)	50.00	50.00	50.00	50.00
Osmolarity (mosm)	285.00	285.00	320.00	

Figure 7

Culture Condition	No. of Experiments	No. of Inseminated Oocytes	Percent Two-cell	Percent Blastocyst
<i>Modified HTF, isotonic</i>	5	58	90% (52/58)	87% (51/58)
<i>Modified HTF, hypertonic</i>	9	234	90% (211/234)	86% (202/234)
$[Tre]^{in} = 0.07M$	3	32	78% (25/32)	66% (21/32)
$[Tre]^{in} = 0.15M$	5	69	67% (46/69)	29% (20/69)

Figure 8



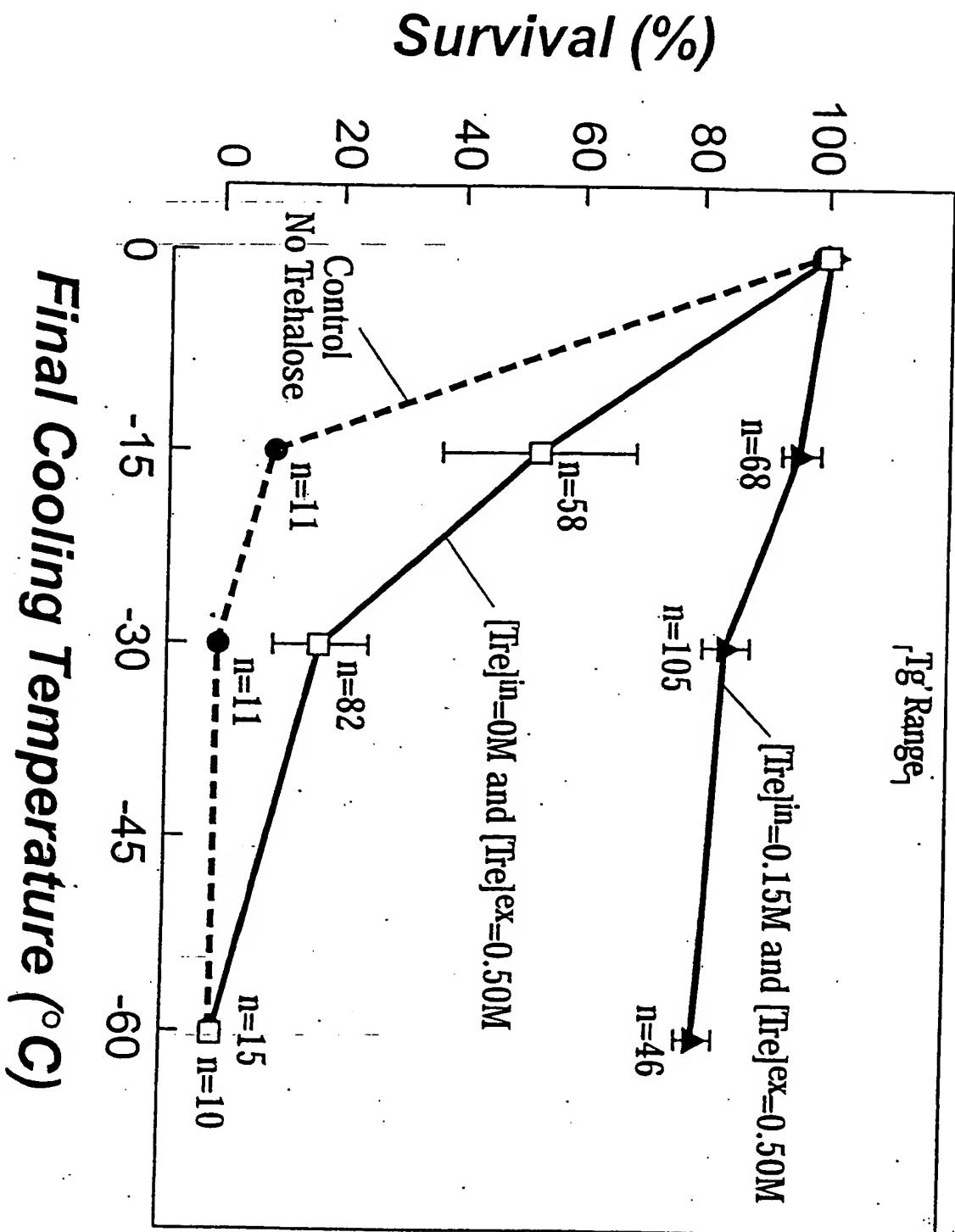


Figure 10

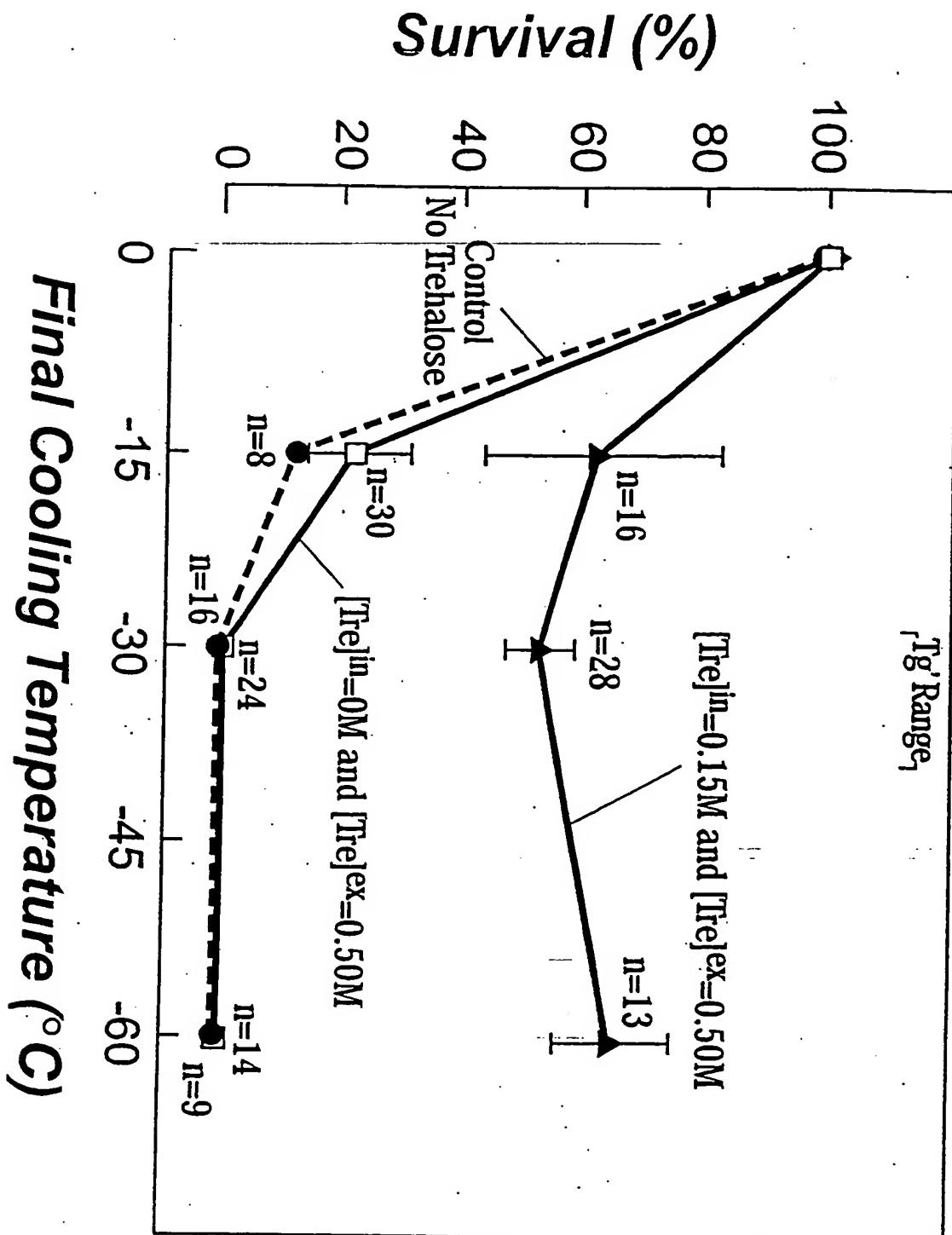
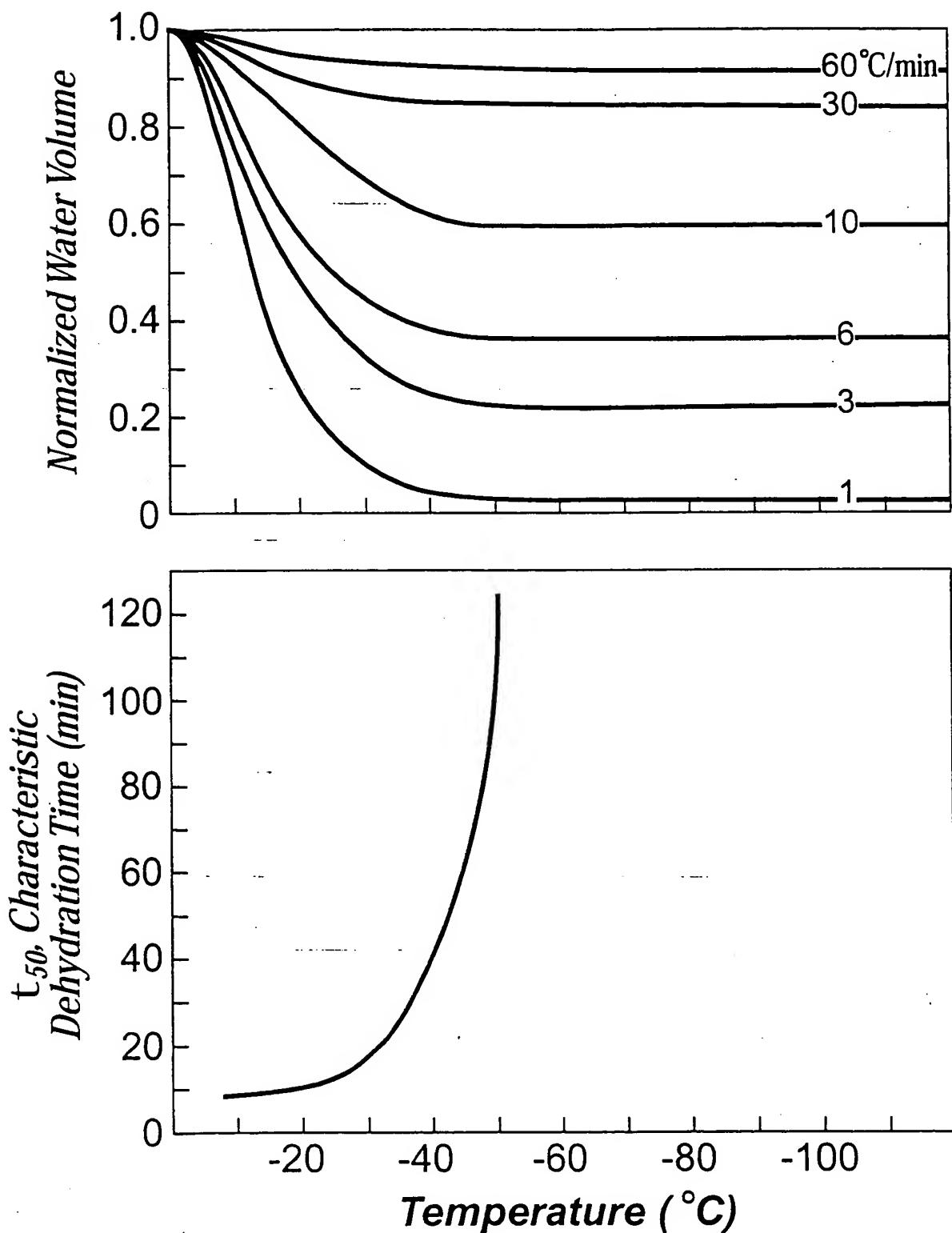


Figure 11



Figures 12A – 12B

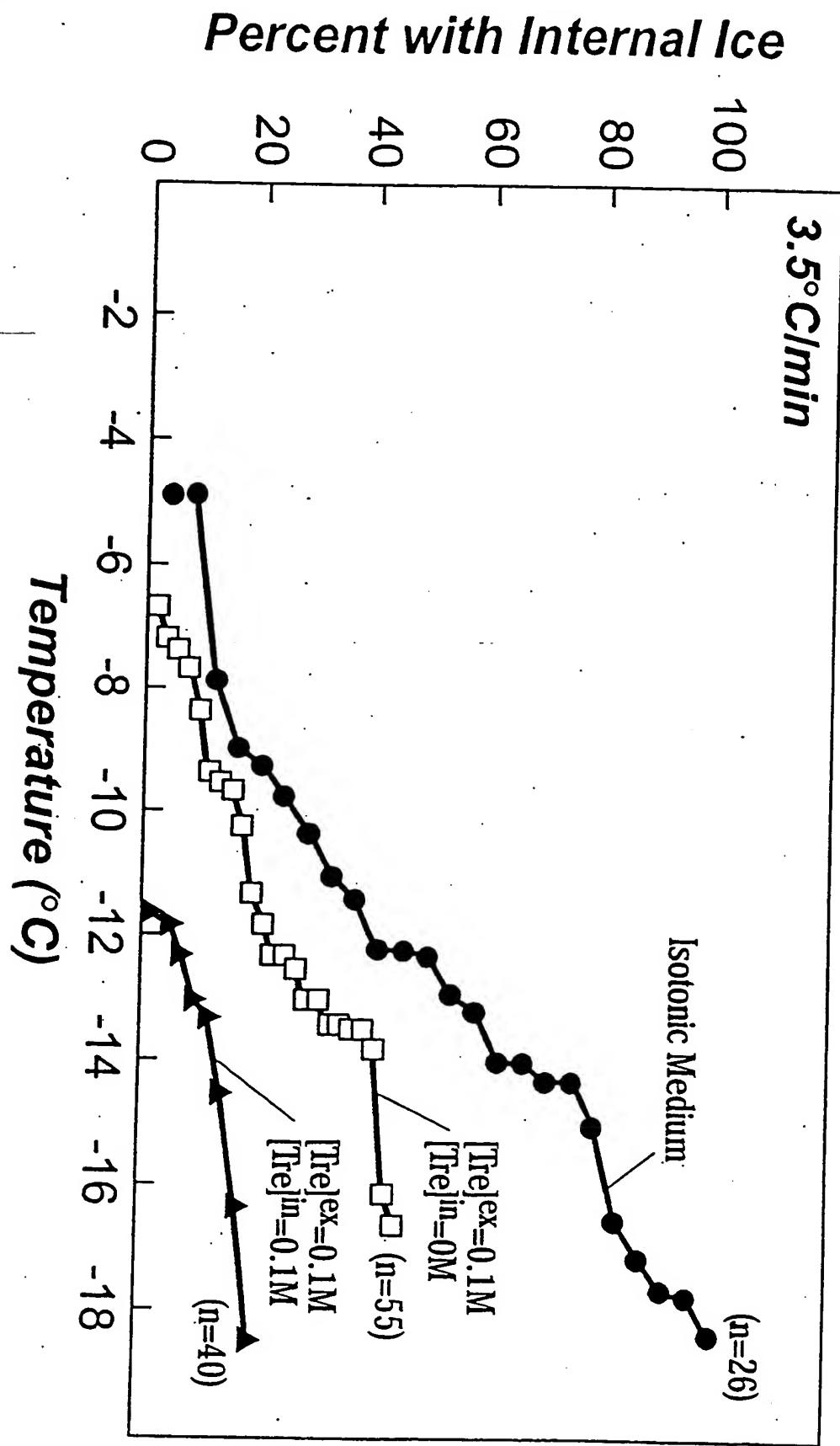


Figure 13

Figure 14

Solute	M _w	T _v °C
erythrose	120.1	-50
threose	120.1	-45.5
erythritol	122.1	-53.5
thyminose (deoxyribose)	134.1	-52
ribulose	150.1	-50
xylose	150.1	-48
arabinose	150.1	-47.5
lyxose	150.1	-47.5
ribose	150.1	-47
arabitol	152.1	-47
ribitol	152.1	-47
xylitol	152.1	-46.5
methyl riboside	164.2	-53
methyl xyloside	164.2	-49
quinovose (deoxyglucose)	164.2	-43.5
fucose (deoxygalactose)	164.2	-43
rhamnose (deoxymannose)	164.2	-43
talose	180.2	-44
idose	180.2	-44
psicose	180.2	-44
altrose	180.2	-43.5
glucose	180.2	-43
gulose	180.2	-42.5
fructose	180.2	-42
galactose	180.2	-41.5

Title: Microinjection of Cryoprotectants For Preservation of
Cells

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Figure 14 Continued

Solute	M _w	T _g °C
allose	180.2	-41.5
sorbose	180.2	-41
mannose	180.2	-41
tagatose	180.2	-40.5
inositol	180.2	-35.5
mannitol	182.2	-40
galactitol	182.2	-39
sorbitol	182.2	-43.5
2-O-methyl fructoside	194.2	-51.5
β-1-O-methyl glucoside	194.2	-47
3-O-methyl glucoside	194.2	-45.5
6-O-methyl galactoside	194.2	-45.5
α-1-O-methyl glucoside	194.2	-44.5
1-O-methyl galactoside	194.2	-44.5
1-O-methyl mannoside	194.2	-43.5
1-O-ethyl glucoside	208.2	-46.5
2-O-ethyl fructoside	208.2	-46.5
1-O-ethyl galactoside	208.2	-45
1-O-ethyl mannoside	208.2	-43.5
glucoheptose	210.2	-37.5
mannoheptulose	210.2	-36.5
glucoheptulose	210.2	-36.5
perseitol (mannoheptitol)	212.2	-32.5
1-O-propyl glucoside	222.2	-43
1-O-propyl galactoside	222.2	-42

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Figure 14 Continued

Solute	M _w	T _g °C
1-O-propyl mannoside	222.2	-40.5
2,3,4,6-O-methyl glucoside	236.2	-45.5
isomaltulose (palatinose)	342.3	-35.5
nigerose	342.3	-35.5
cellobiulose	342.3	-32.5
isomaltose	342.3	-32.5
sucrose	342.3	-32
gentiobiose	342.3	-31.5
laminaribiose	342.3	-31.5
turanose	342.3	-31
mannobiose	342.3	-30.5
melibiose	342.3	-30.5
lactulose	342.3	-30
maltose	342.3	-29.5
maltulose	342.3	-29.5
trehalose	342.3	-29.5
cellobiose	342.3	-29
lactose	342.3	-28
maltitol	344.3	-34.5
isomaltotriose	504.5	-30.5
panose	504.5	-28
raffinose	504.5	-26.5
maltotriose	504.5	-23.5
nystose	666.6	-26.5
stachyose	666.6	-23.5

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Figure 14 Continued

Solute	M _w	T _f , °C
maltotetraose	666.6	-19.5
maltopentaose	828.9	-16.5
α-cyclodextrin	972.9	-9
maltohexaose	990.9	-14.5
maltoheptaose	1153.0	-13.5

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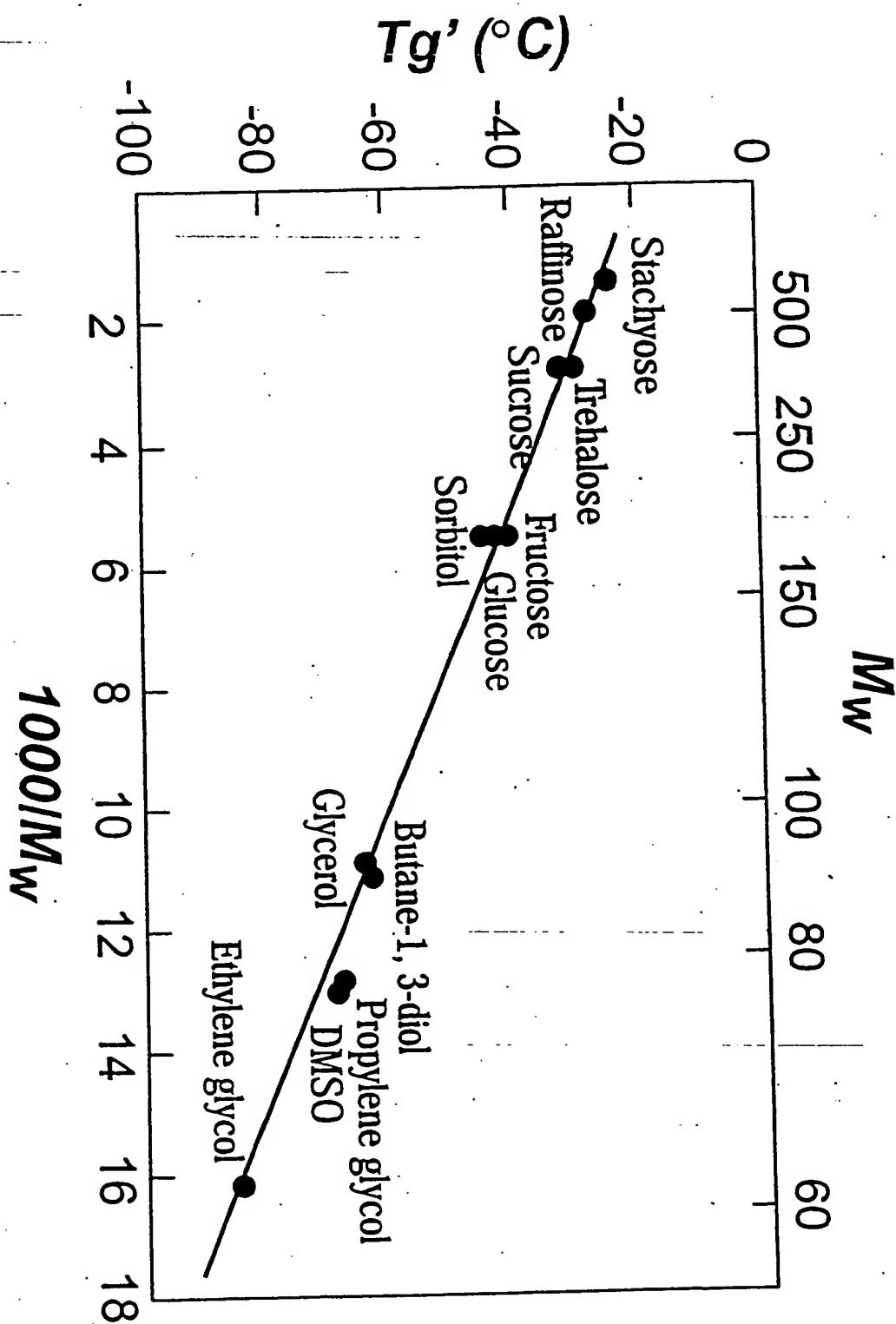


Figure 15

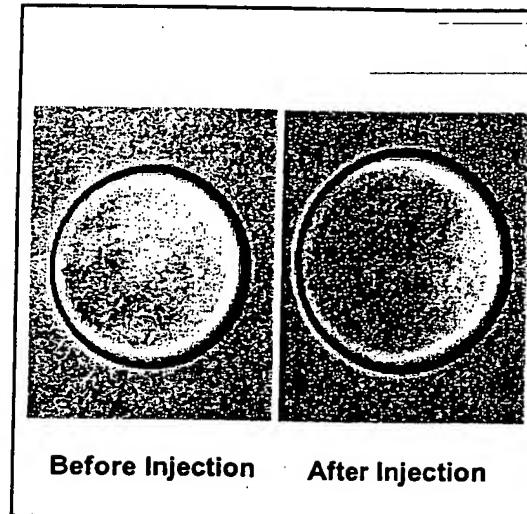


Figure 16

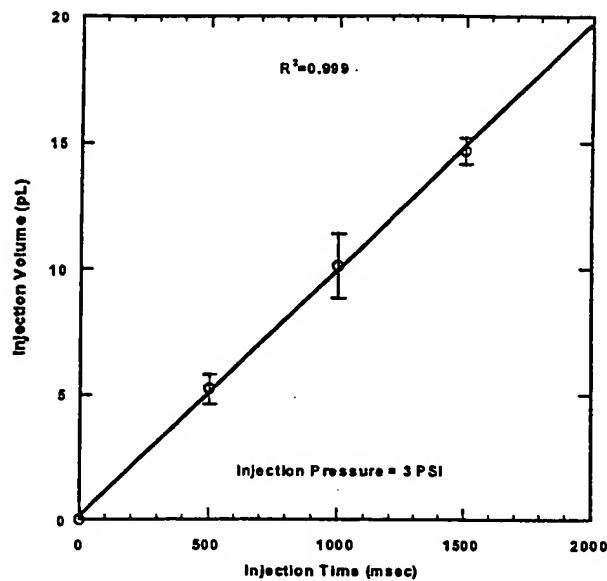


Figure 17

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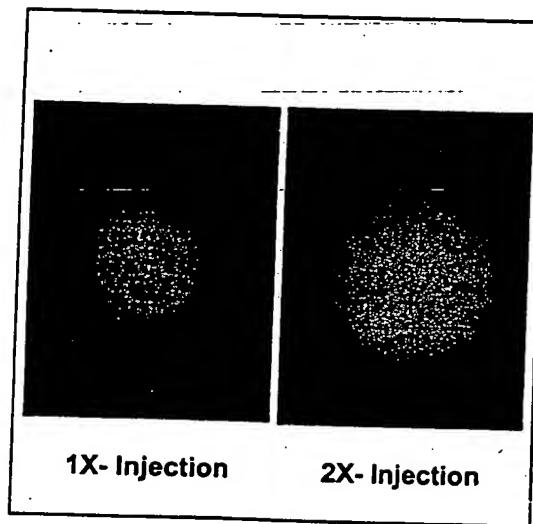


Figure 18

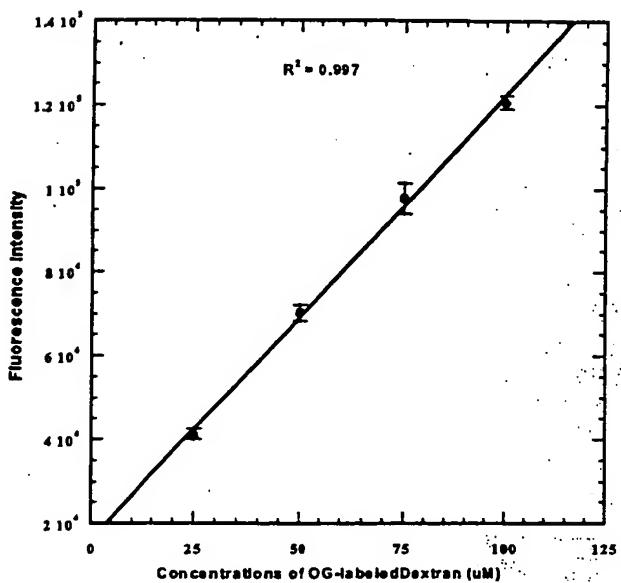


Figure 19

Component	Hyper 320	Hyper 340	Hyper 360
NaCl (mM)	107.02	115.90	123.50
KCl (mM)	5.38	5.83	6.21
KH ₂ PO ₄ (mM)	0.43	0.46	0.49
MgSO ₄ • 7H ₂ O (mM)	0.23	0.24	0.26
NaHCO ₃ (mM)	28.16	30.50	32.50
CaCl ₂ • 2H ₂ O (mM)	2.25	2.44	2.60
Lactate (mM)	22.53	24.40	26.00
Pyruvate (mM)	0.37	0.40	0.43
D- Glucose (mM)	3.13	3.39	3.61
L-Glutamine (mM)	1.13	1.22	1.30
EDTA (mM)	0.01	0.01	0.01
Phenol red (mM)	0.03	0.03	0.03
BSA (mg/mL)	4.00	4.00	4.00
Gentamicin (mg/ml)	50.00	50.00	50.00
Essential amino acids ⁱ (mL/L)	10.00	10.00	10.00
Non-essential amino acids ⁱⁱ (mL/L)	5.00	5.00	5.00
Osmolality	320	340	360

Figure 20

ⁱ [50X Solution, Gibco]

ⁱⁱ [100X Solution, Gibco]

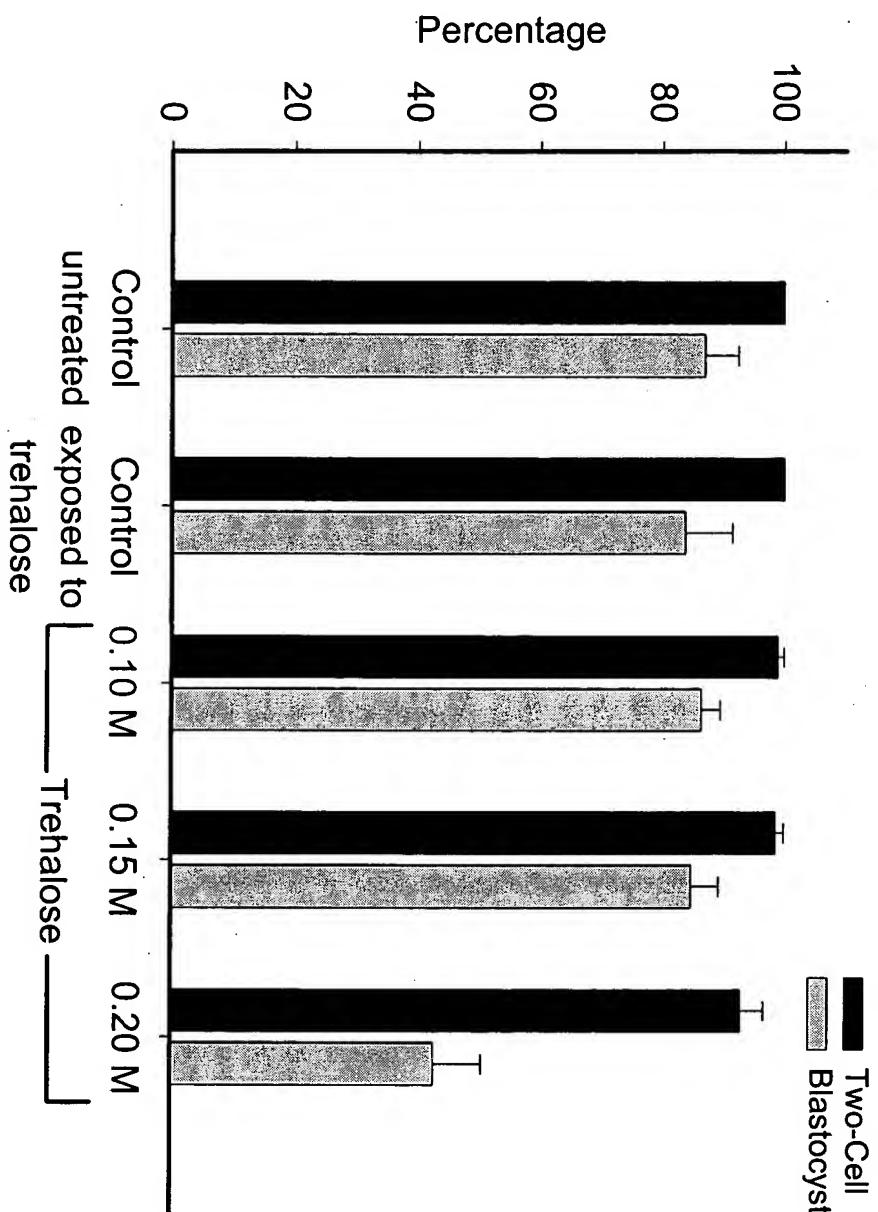


Figure 21